PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

(Chapter II of the Patent Cooperation Treaty)

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference P64870PC00	FOR FURTHER ACTION	See Form PCT/IPEA/416
International application No. PCT/NL2004/000615	International filing date *(day/month/year)* 03.09.2004	Priority date *(day/month/year)* 03.09.2003

International Patent Classification (IPC) or national classification and IPC
B01D25/21, B01L3/00, G01N33/543

Applicant
CEDI DIAGNOSTICS B.V. et al.

1. This report is the international preliminary examination report, established by this International Preliminary Examining Authority under Article 35 and transmitted to the applicant according to Article 36.
2. This REPORT consists of a total of 6 sheets, including this cover sheet.
3. This report is also accompanied by ANNEXES, comprising:
   a. ☒ *sent to the applicant and to the International Bureau)* a total of 4 sheets, as follows:
      ☒ sheets of the description, claims and/or drawings which have been amended and are the basis of this report and/or sheets containing rectifications authorized by this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions).
      ☐ sheets which supersede earlier sheets, but which this Authority considers contain an amendment that goes beyond the disclosure in the international application as filed, as indicated in item 4 of Box No. I and the Supplemental Box.
   b. ☐ *(sent to the International Bureau only)* a total of (indicate type and number of electronic carrier(s)) , containing a sequence listing and/or tables related thereto, in computer readable form only, as indicated in the Supplemental Box Relating to Sequence Listing (see Section 802 of the Administrative Instructions).

4. This report contains indications relating to the following items:

☒	Box No. I	Basis of the opinion
☐	Box No. II	Priority
☐	Box No. III	Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
☐	Box No. IV	Lack of unity of invention
☒	Box No. V	Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
☐	Box No. VI	Certain documents cited
☐	Box No. VII	Certain defects in the international application
☒	Box No. VIII	Certain observations on the international application

Date of submission of the demand	Date of completion of this report
01.07.2005	24.01.2006

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INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

International application No.
PCT/NL2004/000615

Box No. I Basis of the report

1. With regard to the **language**, this report is based on the international application in the language in which it was filed, unless otherwise indicated under this item.

   ☐ This report is based on translations from the original language into the following language , which is the language of a translation furnished for the purposes of:
   - ☐ international search (under Rules 12.3 and 23.1(b))
   - ☐ publication of the international application (under Rule 12.4)
   - ☐ international preliminary examination (under Rules 55.2 and/or 55.3)

2. With regard to the **elements*** of the international application, this report is based on *(replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report)*:

Description, Pages	
1-4, 6-23	as originally filed
5	received on 04.07.2005 with letter of 01.07.2005
Claims, Numbers	
1-14	received on 20.12.2005 with letter of 20.12.2005
Drawings, Sheets	
1/2, 2/2	as originally filed

☐ a sequence listing and/or any related table(s) - see Supplemental Box Relating to Sequence Listing

3. ☐ The amendments have resulted in the cancellation of:
   - ☐ the description, pages
   - ☐ the claims, Nos.
   - ☐ the drawings, sheets/figs
   - ☐ the sequence listing *(specify)*:
   - ☐ any table(s) related to sequence listing *(specify)*:

4. ☐ This report has been established as if (some of) the amendments annexed to this report and listed below had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).
   - ☐ the description, pages
   - ☐ the claims, Nos.
   - ☐ the drawings, sheets/figs
   - ☐ the sequence listing *(specify)*:
   - ☐ any table(s) related to sequence listing *(specify)*:

* *If item 4 applies, some or all of these sheets may be marked "superseded."*

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

International application No.
PCT/NL2004/000615

Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	3
	No:	Claims	1-2, 4-14
Inventive step (IS)	Yes:	Claims	
	No:	Claims	1-14
Industrial applicability (IA)	Yes:	Claims	1-14
	No:	Claims	

2. Citations and explanations (Rule 70.7):

see separate sheet

Box No. VIII Certain observations on the international application

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:

see separate sheet

Re Item V

Reasoned statement with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

Reference is made to the following documents:

D1: US 2003/113713 A1 (WOHLSTADTER JACOB N ET AL) 19 June 2003 (2003-06-19)

D2: WINKLHOFER K F ET AL: "A sensitive filter retention assay for the detection of PrP<Sc> and the screening of anti-prion compounds" FEBS LETTERS, ELSEVIER SCIENCE PUBLISHERS, AMSTERDAM, NL, vol. 503, no. 1, 10 August 2001 (2001-08-10), pages 41-45

D3: BARNETT G R ET AL: "AN IMPROVED MEMBRANE-FILTRATION ENZYME IMMUNOASSAY FOR THE RAPID SEROLOGICAL DIAGNOSIS OF VIRAL INFECTIONS" JOURNAL OF VIROLOGICAL METHODS, vol. 20, no. 4, 1988, pages 323-332

D4: US-A-5 279 937 (ROWE GERALD E) 18 January 1994 (1994-01-18)

D5: GUERIN-MARCHAND CLAUDINE ET AL: "DMISA (dissociated membrane immunosorbent assay), a new ELISA technique performed with blotted samples" JOURNAL OF IMMUNOLOGICAL METHODS, vol. 167, no. 1-2, 1994, pages 219-225

D6: STYA M ET AL: "DOT-BASED ELISA ENZYME-LINKED IMMUNOSORBENT ASSAY AND RIA RADIOMMUNOASSAY 2 RAPID ASSAYS THAT SCREEN HYBRIDOMA SUPERNATANTS AGAINST WHOLE LIVE CELLS" JOURNAL OF IMMUNOLOGICAL METHODS, vol. 73, no. 1, 1984, pages 75-81

1. Article 33(2) PCT

Claims 1-2 and 4-14 are not novel in the sense of Article 33(2) PCT.

D1 (fig. 10C and D) discloses a microtiter plate, where each well of the microtiter plate contains a cluster of fluid containment regions (fig. 10C, 1141), which correspond to the individual containers in the present application.

Said microtiter plate (support) may be rigid and made out of glass, acetate or polystyrene (D1, par 100).

The bottom of said microtiter plate may be non-porous, however for certain applications

also porous (D1, par 112). D1 lists as applications, where filtration membranes as plate bottoms are considered advantageous, assays that employ filtration of solutions through the plate bottom aiming at increasing the mass transport to the plate bottom (faster binding and shorter incubation times) and remove liquid from the well, the latter resulting in less waste fluids, rendering **claim 1** not novel.
Claim 2 is anticipated by D1 (par 68, last 4 lines), describing the arrangement of the discrete assay domains (corresponding to the containers in the present application) within the wells (corresponding to the clusters in the present application).
Example 4 renders **claims 4, 6 and 7** not novel, whereas D1, claim 46 anticipates present **claim 5.**
Claims 8 and 9 is not novel due to D1, par 130, mentioning prions as analytes to be detected.
Claims 10-14 are considered as not novel on grounds of D1, example 4 in combination with par 112, and par 130 in case of claim 12.

2. Article 33(3) PCT

The present application moreover does not meet the criteria of Article 33(1) PCT, because the subject-matter of **claim 3** does not involve an inventive step in the sense of Article 33(3) PCT, since it relates to a multiwell plate as anticipated by D1 (fig. 10C and D in combination with par 112) using a PVDF membrane as plate bottom. Since D1 (par 112) explicitly foresees filtration membranes as plate bottoms, a PVDF membrane is not considered inventive, because it is a well-known filtration membrane.

3. Article 33(4) PCT

Claims 1-14 are industrially applicable.

Re Item VIII
Certain observations on the international application

Article 6 PCT

The application does not meet the requirements of Article 6 PCT, because **claims 3, 8-9**

and 12 are not clear, since the abbreviations "PVDF","BSE", "Sc", "CWD", "CJD" and "TSE" are ambiguous, have no well-recognized standard meaning and should therefore be replaced by the full terms.

applied sample fluid will flow or spill from said first container into an adjacent second container until the fluid level is again even with the container dividing wall or below the position of the passageway. Now when an amount of fluid is applied that is, due to its volume, capable of dividing evenly over all containers within a cluster, then all said containers within that cluster may be provided with an equal volume of sample fluid in a single sample application step.

Preferably, the container dividing walls have a particular minimal height or the position of the passageway between adjacent and connected containers is elevated above the bottom of the container that an amount of sample fluid can be contained therein. Essentially said amount is sufficient for the performing the detection assay on the analyte. A typical dimension of a container is one that is capable of containing, or has a capacity before spill-over, of between 1 and 5000 µl, preferably from 5 to 1000 µl, more preferably between 5 and 250 µl of fluid. Thus, the microtiter plate of the present invention is characterised in the presence of cluster-dividing walls and container-dividing walls, wherein the container-dividing walls allow for spill-over of sample fluid from one container to another. Essentially in a method of the invention, spill-over will occur when the amount of fluid loaded into a container exceeds the capacity before spill-over, also termed herein the spill-over volume. The height of a container-dividing wall is typically 0.1 to 20 millimeters, preferably 1 to 5 millimeters. The height of a cluster-dividing wall is typically 0.1 to 15 millimeters higher than the container-dividing wall, preferably 1 to 5 millimeters higher.

The container dividing and cluster dividing walls may be of any material and may for instance be all clear, white, black or transparent or light-blocking and may in principle be of any color. Preferably, the walls are light-blocking.

A method according to the present invention may in principle be performed for detecting analytes in any liquid sample and in any fluid, such as cell culture supernatants, water (including potable, cooling tower, waste and

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Claims

1.

1. A microtiter plate comprising a plurality of containers of a rigid material selected from the group consisting of glass, polystyrene, polyacryl, polyamide, polyethylene, polypropylene, acrylate butadiene styrene (ABS), Barnox, PVC, nylon, EVA, PET and combinations thereof, wherein the bottom of each container comprises a (semi-)permeable membrane filter capable of directly or indirectly binding an analyte, and wherein each container is separated from an adjacent container by a container dividing wall, wherein the containers are grouped in one or more clusters, each cluster comprising at least two containers, wherein said clusters are separated from adjacent clusters by a cluster dividing wall and wherein at least part of the container dividing wall is lower than the cluster dividing wall or wherein the container dividing wall contains at least one passageway connecting at least two adjacent containers within a cluster, said passageway being at a distance from the bottom of the container and at least partly below the top of the container.

2. Microtiter plate according to claim 1, wherein each cluster of containers comprises at least n^2 containers, wherein n is an integer, preferably an integer from 2-10, more preferably 2-5.

3. Microtiter plate according to claim 1 or 2, wherein said membrane filter comprises PVDF.

4. Microtiter plate according to any one of claims 1 to 3, wherein at least one container in a cluster of containers comprises a capture ligand for specifically binding an analyte to the membrane filter of said container.

5. Microtiter plate according to any one of claims 1 to 4, wherein at least two container in a cluster of containers comprise a different amount of capture ligand for specifically binding an analyte to said membrane filter.

6. Microtiter plate according to any one of claims 1 to 5, wherein at least two container in a cluster of containers comprise a different capture ligand for specifically binding an analyte to said membrane filter.

7. Microtiter plate according to any one of claims 4 to 6, wherein said analyte is an infectious disease agent or an antibody there against.

8. Microtiter plate according to any one of claims 1 to 7, wherein at least one cluster comprises capture ligands specific for the detection of the causative agent of scrapie, BSE, chronic wasting disease and/or Creutzfeldt-Jakob disease.

9. Microtiter plate according to claim 8, wherein at least one cluster comprises capture ligands for the detection of prions PrP^{Sc}, PrP^{BSE}, PrP^{CWD} and/or PrP^{CJD}.

10. A method for the detection of one or more analytes in a liquid sample comprising:

a) providing a microtiter plate according to any one of claims 1 to 9;

b) applying said liquid sample to at least one cluster of containers, filtering said sample through said membrane filter, thereby binding said one or more analytes to said membrane filter or capture ligand, and optionally performing washing steps;

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c) detecting said bound one or more analytes in said containers by performing a binding assay on said membrane filter, said binding assay preferably being a chemiluminescent immunoassay.

11. Method according to claim 10, wherein said one or more analytes comprise an infectious disease agent or an antibody there against.

12. Method according to claim 11, wherein the infectious disease agent is a prion, preferably a TSE-causing prion.

13. Use of a microtiter plate as defined in any one of claims 1 to 9, for detection of analytes in a liquid sample.

14. Use according to claim 13, wherein said detection comprises the simultaneous detection of multiple analytes in said sample.